Oil Spill Eater II AN URGENT CALL FOR CHANGE IN U.S. OIL SPILL RESPONSE

OSE II- a Non-Toxic superior clean up for Dispersant Side-Effects

With Oil Spill Eater II (a one of a kind bioremediation enzyme additive agent) bearing out as an effective first response alternative to the use of chemical dispersants, contrasting evidence has become even clearer that dispersant use has become a huge failure. Given the increasing negative reviews and heightened doubts about dispersant after-effects being 'mild', OSE II has been preapproved in 13 additional countries since May of 2012 illustrating that the industry and global response community are turning away from the use of dispersants.

In fact, dispersants now have a reputation in many countries outside of the U.S. for being an inadequate response method with more and more studies showing an end point of enormous natural resource destruction; thoroughly documented by the track records of the Ixtoc, Valdez and now 2010 Deepwater Horizon spills.

After in-depth reviews of lessons learned during the Gulf of Mexico BP oil spill disaster, the U.S. Government Accountability Office, EPA Office of Inspector General and the President's Gulf Oil Spill Commission have called for a critical review of the response and update of the U.S. National Contingency Plan (NCP).

An important aspect of this review calls for stepping back and determining the efficacy of the use of dispersants due to now clearly questionable tradeoffs of this legacy spill response method.
And just recently, in August of 2012, a coalition of U.S. public health, wildlife, and conservation organizations filed a Clean Water Act lawsuit naming the U.S. Environmental Protection Agency (EPA) for failures in making available science based information on the toxicity levels of dispersants listed on the NCP Schedule, allegedly resulting in faulty decision making during the 2010 Gulf spill.

The U.S. EPA is now being pressed upon to find safer response agents to replace outdated plans which have relied on the use of dispersants, particularly Corexit 9527 and 9500. The use of these chemical response agents have now demonstrated to be more toxic than the oil hence violating the U.S. Clean Water Act (CWA). The CWA stipulates that for a response method to be utilized, it must <u>remove</u> oil from the environment which dispersants do not fulfill. Studies now show that use of dispersants in fact prolong the time that oil + chemical dispersants are left lingering in the environment/water column resulting in adverse impacts to flora and fauna for up to 20 to 30 years as was also proven by historical studies of the Ixtoc, and Valdez spills.

Last month, the U.S. EPA Regional Response Team VI which oversees spill response plans in the Gulf Region, sent a request to their Science and Technology Committee to evaluate OSE II as a first response bioremediation agent that actually removes oil from the environment and is non toxic to the marine species and responders.

As part of this review, OSEI Corporation CEO Steven Pedigo with 25 years experience applying bioremediation agents to spills is lending his expertise to the Science Committee assisting to update the NCP *Bioremediation Guidance* not reviewed or updated since 2001. “One of the most glaring omissions in the EPA guidance” said Mr. Pedigo, “is the fact they have never defined or brought forth the mode of action and proper definitions for the three main types of bioremediation: microbes, nutrients, and enzyme/biosurfactants/nutrient activated bioremediation (OSE II). It appears the US EPA as well as the rest of the world, are now looking for alternatives to dispersants since they do not <u>remove</u> oil from the environment, and exacerbate spill problems” he said.

A copy of this new *Types of Bioremediation* guidance document is published in full below to clarify the subject for all industry stakeholders (oil companies, responsible parties and government agencies) engaged in the development of safer response methods and plan revisions for oil spills to minimize natural resource ruin and greatly reduce the cost of oil spill response.

TYPES OF BIOREMEDIATION, CATEGORY DEFINITIONS AND MODE OF ACTION IN OPEN WATER, MARINE AND FRESH WATER ENVIRONMENTS

August 2012

This document supplements NRT, RRT IV and VI Bioremediation Guidance for the NCP, RCP and ACPs. While covering the essential facts about Bioremediation, the NRT and RRT issued bioremediation guidance materials do not adequately differentiate and define the three primary types of bioremediation categories listed on the NCP Product Schedule and their associated modes of action.

It is important to differentiate the three types of bioremediation processes since their efficacy requires precise application parameters which vary in different types of environments. The limitations and decision points on usage have been covered extensively in previously issued materials but require more simplification, hence this guidance has been provided to simplify the decision making processes.

Essential facts stated in the ***May 2000 NRT SCIENCE AND TECHNOLOGY COMMITTEE-Fact Sheet: Bioremediation in Oil Spill Response*** are:

“Several factors influence the success of bioremediation, the most important being the type of bacteria present at the site, the physical and chemical characteristics of the oil, and the oil surface area….

“Effective bioremediation requires that:

(1) Nutrients remain in contact with the oiled material, and
(2) Nutrient concentrations are sufficient to support the maximal growth rate of the oil-degrading bacteria throughout the cleanup operation.”

NCP PRODUCT TYPES LISTED:

The Bioremediation Agent Types listed on the NCP Product Schedule are deliberately designated and appear as follows:

"1. Microbiological Cultures (MC)
2. Nutrient Additives (NA)
3. Enzyme Additive (EA)"

The first type (MC) constitutes a bioremediation process that utilizes non-indigenous (foreign) bacteria. While useful in controlled environments, a prevailing concern with these types of products has been that the introduction of foreign species might cause future problems which may not become apparent for some time. The second type, (NA) is those agents that contain nutrients or fertilizers to support the microorganisms present in the spill environment. Both are designated as not applicable for open water environments. See 2001 EPA Guidance *Guidelines for the Bioremediation of Marine Shorelines and Freshwater Wetlands* which extensively covers the usage of these two product types which need not be repeated here.

On the other hand, the third type is appropriate as a first response tool in open water environments. Bioremediation (EA) Type has evolved in recent years and has been the subject of considerable technological advances with wide applicability for oil spill response in fresh, brackish, marine and open water environments with temperature ranges as low as 28 degrees. The mode of action of this type will be covered in detail here.

IMPORTANT CONTEXT

The reason for oil spill cleanup is to reduce or eliminate the toxic components, thus enabling the survival of fauna and flora including single cell organisms in each niche of the food chain. Although today's dispersants eliminate the visual and other damaging aspects of the spill on the surface, the spill's toxicity problem has remained in the environment and at times been worsened by the addition of further hydrocarbons in dispersants. The goal of the bioremediation process is to convert oil/hydrocarbon based material to CO_2 and water, thereby permanently removing oil/hydrocarbons from the environment and returning the affected spill area to the pre-spill conditions.

Herewith, the three main types of bioremediation are further defined along with their modes of action to help OSC's, federal, state, and local officials as well as responsible parties to understand and make more informed decisions about bioremediation agents when selecting appropriate oil spill response tools.

CATEGORY TYPE ENZYME ADDITIVE (EA)

As covered, while NRT and RRT guidance addresses the (MC) and (NA) bioremediation types extensively in the 2001 *Guidelines for the Bioremediation of Marine Shorelines and Freshwater Wetlands* it does not sufficiently detail the mode of action of *Bioremediation Type EA*. Below are data to remedy this.

ENZYMATIC AGENT DEFINITION:

Bio-catalysts designed to enhance the emulsification and/or solubilization of oil to make it more available to microorganisms as a source of food or energy. These agents are generally liquid concentrates, which may be mixed with surfactants and nutrients that are manufactured through fermentation. This type of agent is intended to enhance biodegradation by indigenous microorganisms.

(EA) TYPE MODE OF ACTION:

Enzyme Additive mode of action is applicable in open/moving water (fresh, salt and brackish), marsh/estuaries, shoreline and soil environments. When applied, the non-toxic converters and bio-surfactants in Bioremediation Agent (EA) Type eliminate the classic appearance of an oil spill by emulsifying and solubilizing the molecular hydrocarbon structure and eliminating the adhesion properties of crude oil. This usually takes place within the first 5 - 30 minutes (depending on temperature). The emulsified oil continues to float near the surface thereby eliminating a secondary impact to the water column and seabed.

With the toxicity and adhesion properties eliminated, wildlife that may come in contact with the broken down hydrocarbons they will not become coated in oil and oil adherence to marsh, shorelines, sands, and manmade structures is eliminated. The flammability is eliminated in a short time (depending on temperature) protecting ports, harbors and drilling rigs from the potential explosion hazards associated with fuel spills.

A further action of bioremediation category EA, (there are numerous enzymes contained in the product's matrices) is that the enzymes then attach themselves to the hydrocarbons with the biosurfactants, developing protein binding sites, that act as a catalyst to speed up the bioremediation process by inducing enhanced indigenous bacteria to utilize the detoxified oil/hydrocarbons as a food source. The EA category also contains properties that cause all the constituents to remain in contact with the spilled oil/hydrocarbons in moving waters.

Over the next few days or weeks (again, depending on temperature), non-toxic nutrients in the Enzyme Additive type rapidly colonize indigenous bacteria to large numbers. The colonized bacteria consume the detoxified hydrocarbon emulsion, digesting the spill to CO_2 and water, thereby permanently removing the oil/hydrocarbons from the environment and resulting in final water clarification. Without category (EA) assistance, this natural process may take up to 20 years based on Ixtoc and the Valdez spill studies.

SHORELINES/MARSHES:

When a spill has already made land fall or contaminated a marsh, category EA can be applied to lift the spill off the marsh grass (or sandy beaches and shorelines), limiting the time the spill can adversely impact these areas. The use of category EA does not deplete the O2 from water since the spill is held on the surface utilizing predominantly atmospheric O2.

With category EA there are no tradeoffs or deleterious effects with this response method. There is no limited window of opportunity for the application of category EA; it can be used in estuaries, in open (salt) water and, moving fresh water in rivers and soil. It is effective as a first response tool and/or when applied days or months after a spill. Category EA can even be applied to oil that is lying on the seabed floor as long as the product can be brought into contact with the oil which will eventually lift it to the surface returning the seabed to pre-spill conditions.

At the date of this writing, there is only one product on the NCP list that falls under this Bioremediation Agent Type EA classification: (B53-EA-OIL SPILL EATER II)

CATEGORY TYPE MICROBIOLOGICAL CULTURE ADDITIVE (MC)

As covered in NRT Science and Technology Guidance;… *Bioaugmentation,* is a process *"in which oil degrading bacteria are added to supplement the existing microbial population.*".

MICROBIAL AGENTS DEFINITION:

Concentrated cultures of oil-degrading microorganisms grown on a hydrocarbon-containing medium that have been air- or freeze-dried onto a carrier (e.g., bran, cornstarch, oatmeal). In some cases, the microorganisms may be grown-up in bioreactors at the spill site. All commercially available agents use naturally-occurring microorganisms. Some agents may also contain nutrients to assure the activity of their microbial cultures. This type of agent is intended to provide a massive inoculum of oil degrading microbes to the affected area thereby increasing the oil-degrading population to a level where the spilled oil will be used as a primary source of food for energy. Microbial agents are designed to enhance the biodegradation of oil at any, location and would be most useful in areas where the population of indigenous oil degraders is small.

(MC) TYPE MODE OF ACTION:

Bioremediation Agent Type (MC) mode of action utilizes non-indigenous bacteria with the objective to digest oil/hydrocarbons to CO2 and water.

Bioaugmentation is considered a '**polishing up'** or '**finishing**' response product in that it cannot be applied to fresh oil because the toxicity levels kill the added oil degrading bacteria.

When non indigenous bacteria are placed on or near weathered oil these bacteria attempt to release enough quantities of biosurfactants to detoxify the spill so the oil-degrading bacteria will not be adversely impacted by the spill's toxicity, enabling them to use the hydrocarbons as a food source.

The oil degrading bacteria (both indigenous and non indigenous) produce enzymes to develop protein binding sites which permits the bacteria to convert the molecular structure of the hydrocarbons for use as a food source. This process requires a protracted amount of time.

While bioaugmented bacteria acclimate to a spill site, the temperature of the water and or environment, the PH, and the available nutrients, these and other associated and variable environmental conditions may produce adversity that cannot be overcome. These factors along with the unknown time frames associated with their acclamation process are at least partially responsible for the past uncertainty associated bioremediation (MC) type as a viable cleanup methodology.

The application of non-indigenous bacteria generally must be performed where there is very little water movement. Water movement causes the products to dilute to ineffective levels that are unable to stave off the natural competition from indigenous bacteria, and, thus, will not be in sufficient population numbers to produce enough biosurfactants and enzymes to start the breakdown of the molecular structure of the hydrocarbons for a food source. (Lab environments do not emulate this competitive environment; hence, particularly in any area of moving waters, the final outcomes are often uncertain.)

Next to the toxicity of the spill, the most difficult aspect of utilizing non-indigenous bacteria in a foreign environment is the natural competition from the indigenous bacteria that are already acclimated to the spill area; thus, they generally win out.

Bioaugmented bacteria developed specifically for fresh water must be used in fresh water settings only. Products containing salt water bacteria can only be utilized in salt water. (MC) Type is best used on closed and/or controlled environments and is not effective in open water environments.

The use of non indigenous bacteria in most countries is not permitted due to the uncertain effects of allowing non indigenous species in sensitive habits and environments.

CATEGORY TYPE NUTRIENT ADDITIVE (NA)

As covered in NRT Science and Technology Guidance;. . . this next category (NA)--biostimulation is a process *"in which nutrients, or other growth limiting substances, are added to stimulate the growth of indigenous oil degraders*."

NUTRIENT AGENTS DEFINITION:

Bioremediation Agents containing nitrogen and/or phosphorous as the primary means to enhance the rate of growth of indigenous oil-degrading microorganisms. This type of agent is intended to increase the oil-degrading biomass already present in an affected area to a level where the oil will be used as a primary source of food or energy. Because the natural environment may not have sufficient nutrients to encourage bacterial metabolism and growth, extra nutrients may be required. The purpose of this type of agent, therefore, is to provide the nutrients necessary to maintain or increase microbial activity and the natural biodegradation rate of spilled oil.

(NA) TYPE MODE OF ACTION:

The (NA) mode of action involves the general use of nutrients or fertilizers that contain various volumes of Nitrogen N and phosphorous P. The nutrients are placed in conjunction to a spill, where they are expected to enhance the growth and colonization of indigenous bacteria. These bacteria need time to secrete biosurfactants to attack the molecular structure of the spill by solubilizing the oil/hydrocarbons, then emulsifying the spill, increasing the oil-water interface to detoxify the hydrocarbons to the point the enhanced indigenous bacteria can utilize the spill as a food source.

It can be very difficult to apply nutrients or fertilizer in a spill area with toxic oil and still be able to enhance bacteria. Much of the indigenous bacteria are destroyed by the toxicity of the spill initially. Because of the toxicity of the oil, this situation usually precludes the nutrients or fertilizer being capable of enhancing what is left of the indigenous bacteria.

It is also challenging to supply nutrients or fertilizers in a concentration to enhance bacteria without increasing the nitrogen levels to the point that it becomes deadly toxic to aquatic life. An additional problem is getting the nutrients or fertilizers to stay with the oil especially on or in moving waters.

The process of enhancing indigenous bacteria with nutrients and fertilizer and waiting for them to secrete biosurfactants and enzymes in order to start the bioremediation process takes a protracted period of time making (NA) type inappropriate as a first response agent Bioremediation category (NA) can be effectively used where there is little tidal flush, and where the oil has weathered so its toxicity is reduced to the point that indigenous bacteria can survive. This requires NA to be used only as a polishing up agent, with limited scope.

A BRIEF NOTE ON PHYTOREMEDIATION

Phytoremediation has been defined as the use of green plants and their associated microorganisms to degrade, contain, or render harmless environmental contaminants. Phytoremediation of petroleum hydrocarbons generally involves three major mechanisms: (1) degradation, (2) containment and (3) the transfer of contaminants from soil to the atmosphere.

For further information on applicability consult page 87 of http://www.epa.gov/osweroe1/docs/oil/edu/bioremed.pdf

SUMMARY

The three types of bioremediation and their mode of actions as described above have been detailed here to help responders understand how these agents will interact with a spill. The different types and their mode of actions are clearly independent of each other, even though their end point in principle is the same; the ability to reach that end point, and the amount of time it takes to do so, is clearly different.

i Bioremediation [Types MC and NA] for open water spills is not considered to be appropriate or achievable because of the above two requirements. When nutrients are added to a floating slick, they immediately disperse into the water column, essentially diluting to background levels. [with the exception of NCP Listed Type EA based on extensive field use and testing on fresh and weathered hydrocarbons/oil. It recently demonstrated an 80% rate of PAH degradation on Macondo Block La. sweet crude containing Corexit per March 3 2011- BP BCST D.Tsao , LSU R..J. Portier, L. M. Basirico Laboratory Screening of Commercial Bioremediation Agents for the Deepwater Horizon Spill Response.]

ii 2001 Guidelines for the Bioremediation of Marine Shorelines and Freshwater Wetlands (http://www.epa.gov/osweroe1/docs/oil/edu/bioremed.pdf)

iii This description of the EA Type mode of action is based on the NCP listed sole sourced product Oil Spill Eater II's field use and test documentation on fresh and weathered hydrocarbons/oil in ocean, fresh water and shoreline environments. If another EA Type product is added to the NCP List, these descriptions may not apply and should be validated in field tests with that product.

iv As per NRT Science and Technology Committee Bioremediation Fact Sheet: "Added bacteria seem to compete poorly with the indigenous population." … "and has not been shown to have any long-term beneficial effects in shoreline cleanup"

References:

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3. U.S. EPA (2012) NCP Product Schedule,
http://www.epa.gov/oilspill
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EA Type References:

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For U.S. Department of the Interior Bureau of Ocean Energy Management, Regulation and Enforcement (BOEMRE) Herndon, VA; By Louisiana State University

Department of Environmental Sciences http://www.bsee.gov/uploadedFiles/BSEE/Research_and_Training/Technology_Assessment_and_Research/aa%283%29.pdf

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increasing the oxygen take up. http://osei.us/tech-library-pdfs/2011/11-OSEI%20Manual_SaltWaterEfficacyTest.pdf

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PAH's (54% reduction in 30 days on the Maya crude, and medium crude reduced 80% in 30 days.)

20. Environmental Protection Authority New Zealand, Hazardous Substances Division, Benjamin Sowman, (16 July 2012); SOS # 1001797; Determination of the Status of Oil Spill Eater II-Non Hazardous

21. Bioremediation Guidance Appendix A1 Bioremediation Agent EA Type Salt/Fresh Water Toxicity Tests

APPENDIX A1

Bioremediation Agent EA Type Salt Water Toxicity Tests

(EPA standard for toxicity of >100ppm = practically non-toxic)

1. Hap Prichard, OSE II LC50 test 96hr mysid (LC50 of >1900) and 7 day mysid chronic (LC50 of 2500) tests, EPA/NETAC bioremediation protocol development, administrator for NETAC Tom Merski (1992), EPA Research and Development Cincinnati Ohio, http://osei.us/tech-library-pdfs/2011/18-OSEI%20Manual_SaltWaterMarineToxTests.pdf

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>5700, 96hr >5700, 7 day 2.500, Menidia beryline static LC50 48hr 8839, and 96hr 8839mg/l.

3. Enviro Systems Division Resource Analysts Inc New Hampshire Batch 329, OSE II toxicity
test Mysidopsis Bahia LC50 96hr 2100mg/l, March 9 1990
http://osei.us/tech-library-pdfs/2011/18-OSEI%20Manual_SaltWaterMarineToxTests.pdf

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24 Hr >100, 48hr LC50 >100mg/l, fuel oil 48hr LC50 12.6 mg/l, OSE II and Fuel oil 48hr
LC50 29.4mg/l, October 1990
http://osei.us/tech-library-pdfs/2011/18-OSEI%20Manual_SaltWaterMarineToxTests.pdf

EA Type Fresh Water Toxicity Test

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http://osei.us/tech-library-pdfs/2011/17-OSEI%20Manual_FreshWaterSpeciesToxTests.pdf

7. Huther B, Huther and Associates Denton, Texas, (940) 387-1025, for Kwang Keun Kim
South Korea project 05457, OSE II, Toxicity test Pimephales Promelos (minnows) LC50 5856.34mg/l, (l June 2008)
http://osei.us/tech-library-pdfs/2011/17-OSEI%20Manual_FreshWaterSpeciesToxTests.pdf

8. Huther B, Huther and Associates Denton, Texas (940) 387-1025, for Kwang Keun Kim
South Korea project 0S457, OSE II LC50 Toxicity test Ceridaphnia Dubia (water flea) 24hr LC50

>16000mg/l, (l June 2008)
http://osei.us/tech-library-pdfs/2011/17-OSEI%20Manual_FreshWaterSpeciesToxTests.pdf